

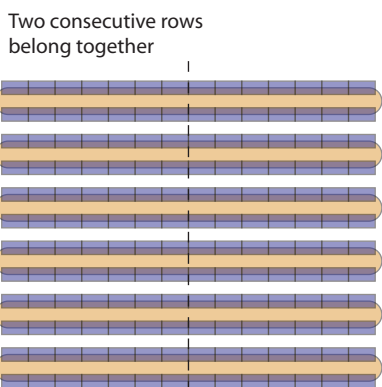
■ One building block (bb)
 height: 2 nm
 length: 16 nt => 1.5 turns => 5.4 nm
 nt = nucleotide (A, T, G, C base)

Inter-helix gay depends on cross-over repetition rate:
 Cross-over every 1.5 bbs => ~ 1 nm gap

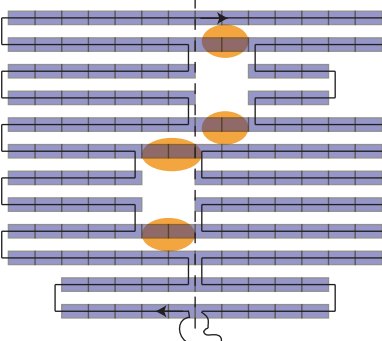
Standard and cheap design: for cyclic, i.e. natural, DNA scaffold
 (e.g. bacteriophage M13mp18 with a length of 7249 basepairs,
 or bacteriophage phiX174 with 5386 bp)

What is a "good" DNA?
 Best is random, as this alleviates the pain of hairpins, where DNA
 within one single strand is complimentary and likes to recombine
 within itself, so those sequences should not be used in the scaffold
 (this is why phiX 174 is better than M13mp18 - it does not have such
 a long hairpin)

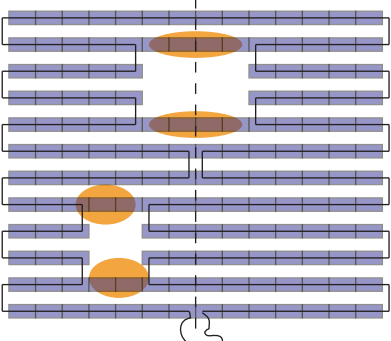
Example bacteriophage DNA - c.f. supplementary material of Rothmund06.pdf



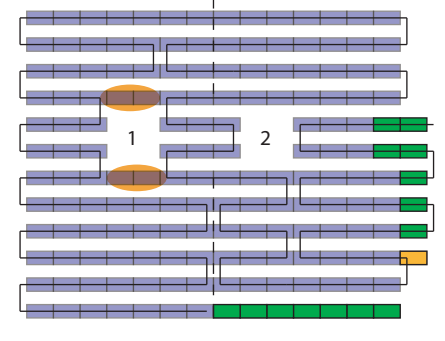
WITH holes at SEAM
 => becomes advanced because
 we have to think of more sophisticated staples
 where the scaffold is missing
 (you will understand later...)



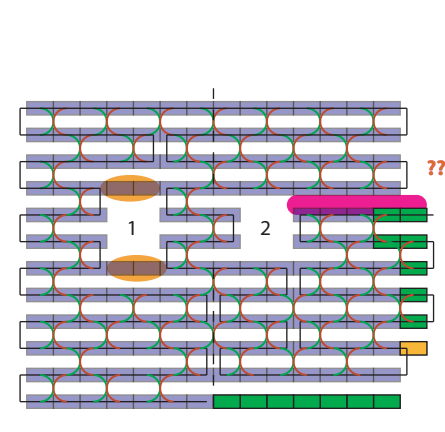
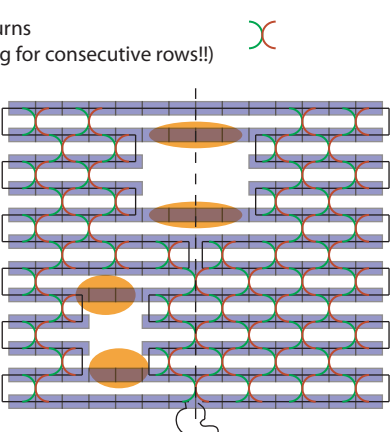
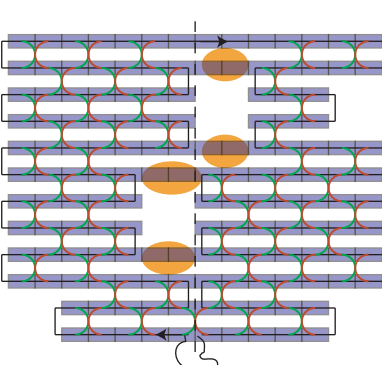
WITH holes not directly next to seam
 => take additional care that you have
 an odd number of bbs in one row
 (This is no big deal as long as there is
 only ONE hole in a pair of rows...)



WITH 2 holes in one pair of row, things
 start getting messy....

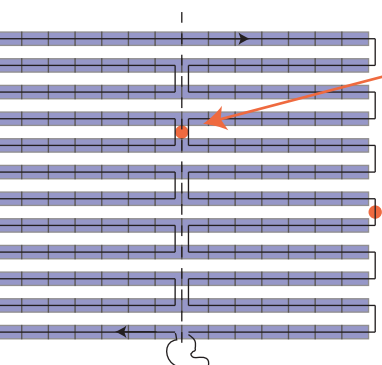


Now start putting the cross-over points every 1.5 turns
 (Within one pair of row - every 3 bbs, but alternating for consecutive rows!!)



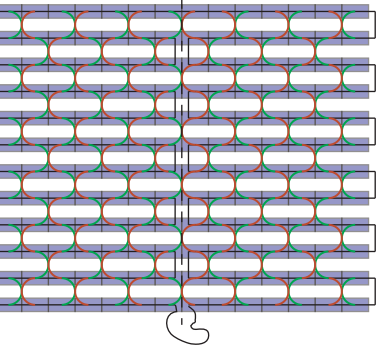
Also, this is not suitable for
 cyclic DNA any more

o.k. ... let's get back to the basic case...

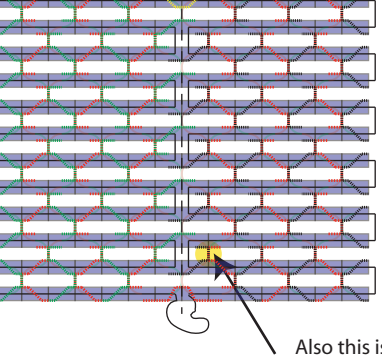


This does NOT represent double stranded (DS) DNA
 later - only for continuous drawing convenience
 (in other words, the vertical scaffold pieces have
 ZERO nt length)

After outlining the scaffold, put the cross-overs



Now put the staples in more detail



Recommended staple length : n x 8 with n = 2,3,4
 The staple cross-connecting the starting and ending point is 48 nt long

— = 16 nt (only extension in x counts)
 ■ = 8 nt (only extension in x counts)

Also this is of ZERO nt length, i.e. not complementary DS DNA

Now ...

- 1) Fill up the scaffold with letters A, T, G, C (if you use natural DNA, you may not choose freely)
- 2) Determine the corresponding staple sequences (A = T and G = C)
- 3) Send your request to a DNA sequencing company
- 4) Mix the scaffold and staple DNA (2 x 50 µl)
- 5) Heat to 92°C, let it cool down to 20°C at a slow cooling rate (~ 0.075°C/min or 4.5 °C/hr) in Thermocycler

{Däumchen dreh' }

- 6) Cleave mica and place 5 µl of DNA solution, let it dry
- 7) Analyse with AFM and pray to find sth resembling your original Design

[Are there possible other analysis techniques, e.g. liquid column chromatography
 to determine molecular weight?]